AMENDMENT TO THE CLAIMS

1. (Currently Amended) A method for analyzing expression frequencies of genes, which comprises the following steps:

(a) ~~a step of~~ forming a vector primer to which each cDNA is ligated, by annealing the vector primer with each mRNA derived from a cell of which expression frequencies of genes is to be analyzed, and synthesizing the cDNA, said vector primer comprising a linear plasmid vector having a single-stranded poly(T) sequence at one 3' end of one strand of the linear plasmid vector, said linear plasmid vector comprising a recognition sequence for a first restriction enzyme ~~at an upstream position from the poly(T) sequence~~, a recognition sequence for a type IIS restriction enzyme, and a recognition sequence for a second restriction enzyme in order from the poly(T) sequence ~~near the 5' end of the strand having the poly(T) sequence, and a recognition sequence for a type IIS restriction enzyme at an downstream position from the recognition sequence for the second restriction enzyme~~, wherein (1) the first restriction enzyme and the second restriction enzyme each digest the vector primer at one position, (2) the cleavage site of the type IIS restriction enzyme is positioned beyond the recognition sequence of the second restriction enzyme, and (3) the vector primer digested with the first restriction enzyme and the type IIS restriction enzyme can be cyclized,

(b) ~~a step of~~ digesting the vector primer to which the cDNA is ligated, with the second restriction enzyme and a third restriction enzyme that does not digest the vector primer and forms a digested end of the same shape as a digested end obtained with the second restriction enzyme, to excise an upstream region of the cDNA, and cyclizing the vector primer,

(c) ~~a step of~~ digesting the cyclized vector primer with the first restriction enzyme and the type IIS restriction enzyme to excise a downstream region of the cDNA so that a tag consisting of a part of the cDNA is left, and cyclizing the vector primer again,

(d) ~~a step of~~ performing polymerase chain reaction (PCR) by using the vector primer as a template and primers to amplify the tag, wherein said primers are oligonucleotides having nucleotide sequences corresponding to known nucleotide regions on each side of the tag that are maintained in the vector primer following digestion in step (c),

(e) ~~a step of~~ ligating the amplification products to form a concatemer of the tags, wherein the tags are separated by known nucleotide sequences introduced by the primers for tag amplification so that no ditags are present in the concatemer, and

(f) ~~a step of~~ determining the nucleotide sequence of the concatemer and investigating types and frequencies of tags occurring in the nucleotide sequence.

2. (Original) The method according to Claim 1, wherein the ligation reaction in the step (e) is performed in the presence of an adaptor having one end of the same shape as an end of the tag to arrange the adaptor at each end of the concatemer, and the concatemer is amplified by performing PCR using an oligonucleotide having a sequence corresponding to the sequence of the adaptor as a primer.

3. (Previously Presented) The method according to Claim 1, wherein, after the step (e), the concatemer is cloned in a cloning vector for nucleotide sequencing, and then the nucleotide sequence of the concatemer is determined.

4. (Previously Presented) The method according to Claim 1, wherein the recognition sequence for the third restriction enzyme consists of four nucleotides.

5. (Previously Presented) The method according to Claim 4, wherein the vector primer has a recognition sequence for a fourth restriction enzyme of which digestion point is in the same position as or a position downstream from the digestion point of the recognition sequence for the second restriction enzyme, which is not excised from the vector primer by the digestion with the type IIS restriction enzyme;

the primer for the downstream side of the tag among the primers used in the step (d) has a recognition sequence for a fifth restriction enzyme that forms an end of the same shape as the end digested with the fourth restriction enzyme; and

the concatemer is formed after the amplified primers are digested with the fourth restriction enzyme and the fifth restriction enzyme.

6. (Currently Amended) The method according to Claim 5, wherein the vector primer has a nucleotide sequence different from the recognition sequence for the fifth restriction enzyme by one nucleotide ~~in~~ at a position upstream from the recognition sequence for the first restriction enzyme, and the nucleotide sequence different by one nucleotide is converted to the recognition sequence for the fifth restriction enzyme by PCR using the primer for the downstream side of the tag.

7. (Original) The method according to Claim 6, wherein the third, fourth and fifth restriction enzymes are identical to one another.

8. (Previously Presented) The method according to Claim 1, wherein the vector primer is formed by ligating a linear plasmid obtained by digesting a plasmid having a multicloning site at two sites in the multicloning site, and a partially double-stranded DNA having an end of the same shape as one end of the linear plasmid and a single-stranded poly(T) sequence.

9. (Previously Presented) The method according to Claim 2, wherein, after the step (e), the concatemer is cloned in a cloning vector for nucleotide sequencing, and then the nucleotide sequence of the concatemer is determined.

10. (Previously Presented) The method according to Claim 1, wherein said vector primer is prepared from a vector selected from the group consisting of pUC18, pUC19,

pBR322, pHSG299, pHSG298, pHSG399, pHSG398, RSF1010, pMW119, pMW118, pMW219, and pMW218.

11. (Previously Presented) The method according to Claim 1, wherein said vector primer is prepared from pUC19.

12. (Previously Presented) The method according to Claim 1, wherein said vector primer is prepared by cloning SEQ ID NO:1 between *Eco*RI and *Hin*dIII within the multiple cloning site of a vector selected from the group consisting of pUC18, pUC19, pBR322, pHSG299, pHSG298, pHSG399, pHSG398, RSF1010, pMW119, pMW118, pMW219, and pMW218.

13. (Previously Presented) The method according to Claim 1, wherein said first restriction enzyme is *Pme*I.

14. (Previously Presented) The method according to Claim 1, wherein said second restriction enzyme is *Bgl*II.

15. (Previously Presented) The method according to Claim 1, wherein said type IIS restriction enzyme is *Bsg*I or *Bsm*FI.

16. (Previously Presented) The method according to Claim 1, wherein said single-stranded poly(T) sequence comprises 10-50 consecutive thymine nucleotides.

17. (Previously Presented) The method of Claim 1, wherein said concatemer comprises 2 to 50 ligated tags.

18. (Previously Presented) The method according to Claim 1, wherein at least one of said primers to amplify the tag is selected from the group consisting of SEQ ID NO:2 and SEQ ID NO:3.

19. (Previously Presented) The method according to Claim 2, wherein the molar ratio of the tags to the adaptor ranges from 1:1 to 1:0.01.

20. (Previously Presented) The method according to Claim 6, wherein at least one of the third, fourth and fifth restriction enzymes is *Mbo*I.

SUPPORT FOR THE AMENDMENT

Claims 1 and 6 have been amended.

Support for the amendment of Claims 1 and 6, can be found in the corresponding claims as originally filed and at page 15, line 3 to page 27, line 2 (for example page 17, lines 1-4 and page 18, lines 4-6).

No new matter has been added by the present amendment.